

## Soil in the agricultural area of Biosphere 2 (1991–1993)

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### Abstract

The agricultural area (intensive agriculture biome) of Biosphere 2 was started with a meter of alkaline soil with high organic content in 1990 and used to support eight persons during closure 1991–1993. Wastes were recycled to maintain fertility. Soils sampled at the end were analyzed for major and minor inorganic constituents and organic matter. C/N ratio was high (13:1 to 17:1). Organic matter decreased 0.2 to 1.4% per year. With exception of high salinity developing in one plot and denitrification in the rice paddy, the soils at the end were functional in supporting crop production. © 1999 Elsevier Science B.V. All rights reserved.

*Keywords:* Biosphere 2; Organic matter; Soil

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### 1. Introduction

This paper describes the starting conditions, management and characteristics of the soils of the agricultural area (intensive agricultural biome, abbreviated IAB) in Biosphere 2 1991–1993. The agricultural area was operated to provide the diet for the eight person crew, who provided the labor. The agriculture provided 80% of their food with 73 g protein, 32 g fat, and 2200 kcal per day (Nelson et al., 1993a,b; Silverstone and Nelson, 1996). Plant, animal and human wastes were returned to

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the soil to recycle nutrients. In March, 1993 soil samples were collected for chemical analysis.

## 2. Plots and soil at the start

The growing area was divided into 18 plots,  $\approx 93 \text{ m}^2$  each. Water was recirculated to two wetland rice paddles which supported a small *Tilapia* population. An orchard area  $172 \text{ m}^2$  contained banana, papaya and understorey fodder crops, partially shaded.

It was decided to use a soil based system rather than a hydroponic or aeroponic system, which eliminated the need for consumables such as nutrient solutions for hydroponics, which could not be produced within the facility or easily reconstituted in appropriate balance and concentration. Using soils took advantage of reasonably stable low-energy, biogeochemical processes which have successfully operated over geologic time-frames in earth's biosphere, providing habitat for the multitude of organisms present in fertile soil that assist in the maintaining of nutrients rather than relying on energy intensive highly technical mechanisms.

The soils used to start the area were high in organic matter. A soil mixture containing 75% clay loam pond soil, 10% Canadian peat moss and 15% compost was installed by hand to a depth of 1 m. This method of placement was used to reduce the risk of compaction. Soil installation and crop production began while construction was still underway in 1990 so that the soils could begin to stabilize. The plots were first planted with a green manure cover crop and later with field crops. By closure in September 1991 there was a 3 month store of food in place. The main agricultural crops were rotated in the 18 plots while the rice paddies produced rice continuously throughout the first two years. Two plots, one of which was sampled and tested in March 1993 (plot 7), were initially planted with *Leuceana leucocephala* trees intended as a perennial fodder crop for the animals. These were removed after 1 year and replaced with other crops.

## 3. Operation

Air handling units were used to regulate temperature and humidity. In winter, temperature ranged between 19 and 27°C and in summer between 19 and 30°C. Relative humidity was kept as low as possible for pest control and generally averaged below 45%.

A subsoil drainage system took leachate water to storage tanks in the basement. Aerobic settling tanks and a wetland wastewater recycling system (Nelson et al., 1999) treated sewage water that was pumped from the human habitat and wash-down water from the barn. Effluent from the wetland was mixed with agriculture leachate and subsequently returned to the soil as irrigation water. Water was condensed from the air in the cooling coils of air handling units, collected and piped to storage tanks in the basement. The leachate, effluent and condensate water

were mixed to obtain a total dissolved solids level of  $\sim 700$  ppm, appropriate for crop irrigation, before returning the water to the agriculture soil. Solid wastes from the animal bay area and from crop residues were broken down using a hammer mill, and during the second year were reincorporated into the topsoil. These nutrient sources were contained or recycled within the IAB with the following exceptions: First, as a portion of the animal fodder (e.g. savannah grasses) it was collected from the wilderness area, an equivalent amount was returned to the wilderness soils and used as mulch. Second, on several occasions the water storage tanks in the agriculture facility became too full—often because the rice paddies had been drained for replanting. Water was then pumped from the agricultural storage tanks into the main storage reservoirs in the south lung. Third, by the beginning of the second experiment the bulk of the unrecycled biomass in the agricultural area were the trunks of the *Leuceana* trees that had been removed from the plots at the end of the first year. These were stored for future recycling.

Artificial lighting was installed during the transition phase between the first and second experiment to add  $\approx 5 \text{ mol m}^2$  per day, which boosted winter light levels from 30 to 50%.

Synthetic insecticides were not used in Biosphere 2 because of the risk to biota and humans within a closed system. Pest management techniques included use of diversified cropping, use of pest-resistant cultivars, introduction of beneficial insects, environmental manipulations, manual intervention and the use of non-toxic sprays (Silverstone and Nelson, 1996).

#### 4. Soil sampling and analysis

The soil samples were collected using standard methods (Peterson and Calvin, 1986), from eight areas: six of the agriculture plots, the rice paddy and the orchard area. Three replicate samples were taken from the north, center, and south of the agricultural plots from the upper soil 0–30 cm deep, each replicate sample consisting of a composite of five separate cores ( $\approx 0.6$  kg of soil per replicate sample). In the orchard the three samples were taken from the east, center and west. One sample core was collected at below 90 cm from each sampling area. For tissue analysis 0.6 kg wet weight of  $\approx 3$  month old leaves were taken from seven of the most important agronomic crops: banana, rice, wheat, *Lablab purpureus*, hyacinth bean, papaya, squash and beet.

Soils were analyzed by standard methods (Page et al., 1982). Soil samples, which arrived at the laboratory cooled and field-moist, were sieved wet and the  $< 2$  mm fraction divided. One portion was kept under refrigeration for microbiological analysis, and the other dried at  $60^\circ\text{C}$  for chemical analysis. Saturated pastes were prepared from the dried soil samples with deionized water, and pH was measured both in the paste and the saturation extract; electrical conductivity was measured in a subsample of the water extract. The concentrations of P, B, Mg, Ca, K, B, Cu, Mn, Zn and Mo, in the soil solution (saturation extract) were measured by direct-current plasma spectrometry (DCP) (SpectraSpan VB, Fisons Instruments, Danvers, Maryland).

Inorganic N (nitrate, nitrite and ammonium) was extracted within 2 days from the moist refrigerated soil with 2 M KCl, and N was measured in the extract by automated colorimetry (QuikChem, Lachat Instruments, Milwaukee, WI). The concentrations of total N, extractable P and exchangeable cations were also measured by colorimetry. Total N in the soil was measured by the Kjeldahl procedure.

Plant-available P was estimated by extracting with 0.5 M NaHCO<sub>3</sub> at pH 8.5 (Olsen method). Cation exchange capacity was determined by equilibrating with 1 N ammonium acetate, washing with ethanol, and displacing adsorbed ammonium with 10% sodium chloride; the displaced ammonium is equivalent to CEC. Exchangeable K, Mg and Ca were measured by extracting with 1 N ammonium acetate.

The concentration of organic matter in the soil was determined by oxidation of organic C with potassium dichromate-concentrated sulfuric acid and titration with ferrous sulfate (Walkley–Black method). Archival soil samples were also obtained and analyzed for organic C to check whether significant losses of organic matter might have occurred since the soil was placed in the IAB. Soil microbial biomass was determined by the chloroform fumigation-incubation method. The 10-day CO<sub>2</sub> flush was measured in alkali traps by automatic double end-point titration. Microbial respiration was measured in the unfumigated controls. Nitrogen in plant tissues was measured in concentrated sulfuric acid digests; other nutrients were determined by DCP in nitric acid digests. Bulk density ( $0.99 \pm 0.11$ ,  $n = 3$ ) was subsequently determined by extracting soil cores to a depth of  $\approx 1$  m in agricultural plots 2 and 17 and orchard.

## 5. Results

Results of soil analysis are given in Tables 1–3. Soils were slightly to moderately alkaline. In most plots, salinity in surface soils (0–30 cm) was at or above the threshold ( $2 \mu\text{S}/\text{dm}$ ) at which sensitive crops may be affected (Table 1) and subsurface soils ( $> 90$  cm) in several plots were saline ( $> 4 \mu\text{S}/\text{dm}$ ). The soil in plot 7 was strongly saline and one sample was also sodic ( $\text{SAR} > 13$ ). The irrigation water, although slightly alkaline, was adequately low in salts and sodicity. Both the irrigation water and leachate were high in inorganic N, and the concentration of ammonium in the deep soil of the orchard approached the level for incipient ammonia toxicity for moderately alkaline soils.

The cation exchange capacity was moderately high ( $27 \pm 6.5$  cmol/kg,  $n = 8$ ), but typical for soils high in organic matter. The concentrations of extractable (e.g. ‘plant-available’) inorganic N (Table 1) and of exchangeable Mg (Table 2) in the IAB surface soils were mostly near the mean for arable soils, but concentrations of extractable P, and of exchangeable K and Ca were generally much higher than the mean values for arable soils (Bradford et al., 1971; Mengel and Kirby, 1979). In the soil solution (e.g. saturation extract), the concentrations of Ca and K were higher,

Table 1  
Chemical and biological properties of soil and water samples from the intensive agriculture biome, Biosphere 2

| Plot                        | Depth | pH saturation extract | Electrical conductivity (dS/M) | Sodium adsorption ratio (SAR) (mol mol <sup>-1/2</sup> ) | Organic matter <sup>a</sup> (%) | Microbial carbon <sup>b</sup> (mg/kg) | Microbial C as % of organic C | Total N (Kjeldahl) (mg/kg) | Total <sup>c</sup> extractable N (mg/kg) | Extractable P (mg/kg) |
|-----------------------------|-------|-----------------------|--------------------------------|--|---------------------------------|---------------------------------------|-------------------------------|----------------------------|--|-----------------------|
| Orchard                     | 0–30  | 7.5                   | 2.0 ± 1.0                      | 2.0 ± 0.6  | 6.66 ± 0.15                     | 647 ± 71.3                            | 1.65 ± 0.14                   | 2667 ± 462                 | 46 ± 36                                  | 102 ± 10              |
|                             | >90   | 7.4                   | 4.0                            | 2.7  | –                               | 631                                   | –                             | 2300                       | 123                                      | 113                   |
| Rice Field                  | 0–30  | 7.7                   | 1.9 ± 0.2                      | 2.1 ± 0.5  | 4.12 ± 0.41                     | 418 ± 99.5                            | 1.71 ± 0.26                   | 1467 ± 115                 | 9.5 ± 3.8                                | 60 ± 8                |
|                             | >90   | 7.6                   | 1.9                            | 1.8  | 4.45                            | 410                                   | 1.57                          | 1600                       | 43                                       | 64                    |
| Plot 2                      | 0–30  | 7.6                   | 1.9 ± 0.2                      | 1.3 ± 0.2  | 4.81 ± 0.24                     | 536 ± 27.2                            | 1.89 ± 0.18                   | 1833 ± 58                  | 62 ± 15                                  | 72 ± 11               |
|                             | >90   | 7.5                   | 2.0                            | 1.5  | 4.71                            | 297                                   | 1.07                          | 2033                       | 84                                       | 73                    |
| Plot 7                      | 0–30  | 7.2                   | 5.9 ± 0.8                      | 7.1 ± 9.9  | 5.55 ± 0.35                     | 476 ± 109                             | 1.47 ± 0.39                   | 2200 ± 100                 | 80 ± 91                                  | 70 ± 15               |
|                             | >90   | 7.2                   | 4.5                            | 1.2  | –                               | 453                                   | –                             | 2200                       | 180                                      | 149                   |
| Plot 8                      | 0–30  | 7.9                   | 2.5 ± 0.7                      | 1.1 ± 0.4  | 5.03 ± 0.26                     | 516 ± 38.6                            | 1.75 ± 0.14                   | 2067 ± 58                  | 149 ± 132                                | 88 ± 10               |
|                             | >90   | 7.0                   | 5.4                            | 1.3  | 5.06                            | 463                                   | 1.56                          | 2500                       | 107                                      | 149                   |
| Plot 12                     | 0–30  | 8.0                   | 2.2 ± 1.0                      | 1.3 ± 0.3  | 6.64 ± 1.05                     | 581 ± 129                             | 1.48 ± 0.14                   | 3067 ± 907                 | 64 ± 62                                  | 128 ± 18              |
|                             | >90   | 7.6                   | 1.4                            | 1.5  | 6.68                            | 609                                   | 1.55                          | 3300                       | 20                                       | 82                    |
| Plot 16                     | 0–30  | 7.8                   | 2.2 ± 0.3                      | 1.6 ± 0.1  | 5.64 ± 0.59                     | 529 ± 146                             | 1.58 ± 0.34                   | 2400 ± 624                 | 76 ± 31                                  | 84 ± 5                |
|                             | >90   | 8.0                   | 1.7                            | 1.5  | 5.90                            | 479                                   | 1.38                          | 1900                       | 87                                       | 78                    |
| Plot 17                     | 0–30  | 7.6                   | 2.7 ± 0.7                      | 1.1 ± 0.5  | 6.37 ± 0.56                     | 669 ± 80.2                            | 1.79 ± 0.14                   | 2800 ± 300                 | 97 ± 49                                  | 103 ± 27              |
|                             | >90   | 7.6                   | 8.1                            | 2.4  | 4.69                            | 469                                   | 1.70                          | 1900                       | 460                                      | 92                    |
| Irrigation H <sub>2</sub> O |       | 7.5                   | 0.7                            | 1.0  |                                 |                                       |                               |                            | 87.3                                     |                       |
| Leachate H <sub>2</sub> O   |       | 8.0                   | 1.8                            | 1.7  |                                 |                                       |                               |                            | 55.4                                     |                       |

<sup>a</sup> Walkley–Black method (assuming 77% oxidation, and assuming soil organic matter consists of 58% organic carbon).

<sup>b</sup> Chloroform fumigation-incubation; 10-day CO<sub>2</sub> flush only, not corrected for unfumigated controls.

<sup>c</sup> (NO<sub>3</sub><sup>-</sup>-N + NO<sub>2</sub><sup>-</sup>-N + NH<sub>4</sub><sup>+</sup>-N).

Table 2

Concentrations of nutrient elements in the soil solution saturation extracts of irrigation and leachate water of the intensive agriculture biome<sup>a</sup>

| Plot                        | Depth (cm) | P         | B         | Mg       | Ca          | K          | Na         | Cu  | Mn  | Zn  | Mo  |
|-----------------------------|------------|-----------|-----------|----------|-------------|------------|------------|-----|-----|-----|-----|
| Orchard                     | 0–30       | 3.2 ± 0.3 | 0.3 ± 0.4 | 33 ± 14  | 214 ± 88    | 111 ± 52   | 121 ± 53   | 0.6 | 0.1 | 0.2 | 0.1 |
|                             | >90        | 5         | 0         | 62       | 2090        | 330        | 380        | 1.1 | 0.7 | 0.1 | 0.2 |
| Rice Field                  | 0–30       | 3.1 ± 1.1 | 0.3 ± 0.1 | 34 ± 11  | 660 ± 740   | 147 ± 30   | 169 ± 70   | 0.8 | 1.8 | 0   | 0.2 |
|                             | >90        | 3         | 0         | 34       | 700         | 127        | 167        | 0.8 | 2.4 | 0   | 0.2 |
| Plot 2                      | 0–30       | 2.7 ± 1.5 | 0.3 ± 0.1 | 29 ± 18  | 285 ± 1070  | 103 ± 47   | 129 ± 60   | 0.7 | 1.7 | 0   | 0.1 |
|                             | >90        | 3         | 0         | 36       | 880         | 103        | 132        | 0.8 | 1.4 | 0   | 0.1 |
| Plot 7                      | 0–30       | 3.2 ± 3.9 | 0.3 ± 0.1 | 35 ± 46  | 859 ± 0     | 102 ± 1108 | 116 ± 1700 | 0.7 | 0.1 | 0   | 0.1 |
|                             | >90        | 2         | 0         | 68       | 2010        | 215        | 207        | 0.6 | 0   | 0.1 | 0.1 |
| Plot 8                      | 0–30       | 0.9 ± 0.1 | 0.0 ± 0.1 | 24 ± 8   | 830 ± 1020  | 72 ± 24    | 94 ± 30    | 0.7 | 0   | 0   | 0.1 |
|                             | >90        | 7         | 0         | 159      | 2120        | 269        | 231        | 0.5 | 0.5 | 0.1 | 0.2 |
| Plot 12                     | 0–30       | 5.0 ± 2.4 | 0.3 ± 0.1 | 53 ± 40  | 790 ± 1150  | 108 ± 70   | 111 ± 72   | 0.7 | 0.1 | 0.1 | 0.1 |
|                             | >90        | 5         | 1         | 39       | 140         | 121        | 80         | 0.5 | 0.5 | 0.1 | 0.3 |
| Plot 16                     | 0–30       | 5.3 ± 1.0 | 0.5 ± 0.4 | 53 ± 11  | 260 ± 80    | 109 ± 23   | 109 ± 22   | 0.7 | 0.2 | 0.1 | 0.1 |
|                             | >90        | 3         | 0         | 23       | 95          | 93         | 62         | 0.3 | 0.7 | 0   | 0.1 |
| Plot 17                     | 0–30       | 3.0 ± 0.3 | 0.3 ± 0.2 | 31 ± 7.6 | 1500 ± 1100 | 132 ± 22   | 125 ± 27   | 0.9 | 0.1 | 0   | 0.2 |
|                             | >90        | 4         | 0         | 173      | 2130        | 2050       | 440        | 0.1 | 0.6 | 0.1 | 0.2 |
| Irrigation H <sub>2</sub> O |            | 1.7       | 0.1       | 8        | 35          | 27         | 24         | 0   | 0   | 0   | 0   |
| Leachate H <sub>2</sub> O   |            | 1.8       | 0.2       | 25       | 130         | 82         | 80         | 0.2 | 0   | 0   | 0   |

<sup>a</sup> Mean mg/l ± S.D.

Table 3

Concentrations of nutrient elements in plant tissues from the intensive agriculture biome

| Crop         | Plot    | Na <sup>a</sup> | P <sup>a</sup> | K <sup>a</sup> | Ca <sup>a</sup> | Mg <sup>a</sup> | Zn <sup>b</sup> | Fe <sup>b</sup> | B <sup>b</sup> | Mn <sup>b</sup> | Cu <sup>b</sup> | Mo <sup>b</sup> |
|--------------|---------|-----------------|----------------|----------------|-----------------|-----------------|-----------------|-----------------|----------------|-----------------|-----------------|-----------------|
| Sweet potato | 8       | 4.1             | 0.5            | 3.5            | 1.8             | 0.5             | 29              | 95              | 63             | 36              | 16              | 9               |
| Banana       | Orchard | 2.7             | 0.2            | 2.7            | 0.3             | 0.3             | 27              | 91              | 38             | 49              | 10              | 4               |
| Rice         | 10      | 2.2             | 0.3            | 1.0            | 0.9             | 0.2             | 15              | 117             | 66             | 330             | 9               | 6               |
| Wheat        | 12      | 4.9             | 0.7            | 2.9            | 0.5             | 0.3             | 30              | 102             | 46             | 22              | 10              | 8               |
| Lablab       | 2       | 4.0             | 0.5            | 2.2            | 4.3             | 0.4             | 35              | 102             | 105            | 22              | 10              | 18              |
| Papaya       | Orchard | 3.5             | 1.1            | 3.1            | 3.6             | 1.7             | 36              | 90              | 356            | 28              | 12              | 16              |
| Squash       | 3       | 5.9             | 0.9            | 3.9            | 2.0             | 0.3             | 41              | 95              | 37             | 15              | 15              | 11              |
| Beets        | 3       | 4.9             | 0.2            | 3.5            | 1.7             | 0.9             | 21              | 77              | 42             | 27              | 9               | 7               |

<sup>a</sup> % Of dry matter.<sup>b</sup> mg/kg dry matter.

P was in the same range or slightly higher, and the toxic elements Na and B were lower than the means for 68 soil samples representing 30 soil series from agricultural areas of California (Bradford et al., 1971). The concentrations of micronutrients in the soil solution were generally near the mean values for arable soils (Bradford et al., 1971). In plant tissues, the concentrations of both macro- and micronutrients were near the means for crop plants that do not show deficiency or toxicity symptoms (Gauch, 1972).

### 5.1. Organic matter

Results of analysis of organic matter are included in Table 1 and Fig. 1. Levels were still high in March 1993. At 1 year prior to closure, organic matter was 6–8% in all plots. But it was still high in the March 1993 samples, having trended downward to 4–6% in the agronomic plots and rice field (Table 1, Fig. 1). The

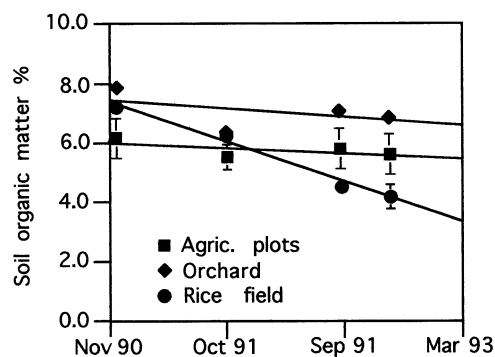


Fig. 1. Dynamics of soils organic matter in the IAB surface soils determined by analysis of archival and March 1993 samples. Error bars are  $\pm 1$  SD. Some data points consist of only a single value. Slopes are 0.2, 0.4 and 1.4% per annum for the agricultural plots, orchard and rice paddy, respectively. Differences in sampling protocols and lack of replication on some dates preclude proper statistical analysis.

orchard, which had the highest organic matter content initially, had lost only about 1% (Fig. 1). Although samples are few, we estimate that annual losses of soil organic matter may be ~0.2, 0.4 and 1.4% in the agronomic plots, orchard and rice field, respectively (Fig. 1). The ratio of organic C to total N ranged from 13:1 to 17:1, higher than typical values 10:1 to 12:1 in humid temperature regions (Brady and Weil, 1996).

## 6. Discussion

The mildly alkaline pH, the build-up of salts in the deep and surface soils, and the development of saline-sodic conditions in plot 7 indicate that the leaching fraction—that is, the extra irrigation water usually added to help flush salts below the root zone—was probably not large enough in the agricultural biome. The higher salinity level in plot 7 was possible due to the production of rapidly growing *L. leucocephala* trees for fodder. These trees required more irrigation than other crops, and higher evapotranspiration from the trees reduced to leachate, probably accounting for the salt build-up. The soil in this plot was heavily irrigated to leach salts, and subsequent tests in September 1993 showed that salinity levels had been reduced.

### 6.1. Nitrogen

Judging from elemental concentrations in soils (Tables 1 and 2) and plant tissues (Table 3), the availability of macronutrients was adequate, and concentrations of potentially toxic chemicals were low. The combination of high inorganic N in the irrigation water and in the soil, however, may have inhibited biological nitrogen fixation and promoted vegetative growth at the expense of fruiting in crops like sweet potato and tomato, which require lower N levels at the onset of fruiting or tuber growth. Concentrations of inorganic N higher than 14 ppm restrict nodulation (Mengel and Kirby, 1979). Observations revealed that there was very little nodulation on legume roots. On the recommendation of crop scientists from Tuskegee University, encouraging tuber formation required ‘dry shocking’ the crop, i.e. not watering it generally for ~4 weeks until plants showed some signs of wilting and early signs of tuber formation were seen on the roots. To encourage tuber formation, vines were also regularly pruned, especially the long runners, and used as fodder for domestic animals. During the second year, an attempt was made to depress N levels and promote tuber growth by incorporating dry uncomposted organic matter with high C:N ratios into the soils, particularly in plots that were going to be planted with sweet potato. Incorporating such residues tends to immobilize N for a time due to microbial demand for N during decomposition (Brady and Weil, 1996).

In the rice field, low inorganic N concentration (Table 1), combined with the apparent high decomposition rate of organic matter (Fig. 1), suggest that significant denitrification was taking place in the flooded soil. A series of rice crops had been



grown in the rice fields, and the soil had been kept continuously flooded for the first 18 months of the experiment. The rice field may thus have been an important source of nitrous oxide to the atmosphere and of organic acids to the leachate. Some of the CO<sub>2</sub> flux to the atmosphere may have originated in the rice paddy through microbial conversion of organic matter to organic acids, which would have been fully oxidized later when the leachate reached a more aerobic environment.

### 6.2. Soil respiration

By 1993 microbial carbon in the agricultural area was low, 1.4–1.89 (Table 1), whereas temperate-zone agricultural soils in carbon equilibrium have C<sub>mic</sub>:C<sub>org</sub> in the range of 2.3 to 2.9 (Anderson and Domsch, 1989).

The excessive carbon dioxide release that generated 600–4500 ppm in the Biosphere 2 atmosphere may have been decreasing. Eventually a steady state balance with the atmosphere could develop.

## 7. Conclusion

Analysis performed after 18 months of operation of the IAB showed that the main problems at the time were high salinity in one of the plots and denitrification in the rice paddies, which had low levels of extractable nitrogen. Otherwise, nutrient recycling techniques and soil management strategies had maintained adequate levels of soil fertility during the 2 years of closure and were not limiting to crop production.

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